A SPOROZOON OF *PHLEBOTOMUS PAPATASII*

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During a routine examination of 1,037 *Phlebotomus papatasii*, of which 939 were females, one female was found to contain oocysts. All the insects were collected in Jericho between April and June, 1925. The infected specimen was one of a number used for experimental purposes and dissected immediately after a feed on a laboratory assistant on 25th May, 1925.

On dissecting the head from the thorax a large number of small glistening bodies were seen to emerge from the thorax and these on examination proved to be sporocysts from a ruptured oocyst.

Further dissection revealed the fact that thorax and abdomen contained four ripe oocysts, one in the thorax and three in the abdomen. The oocysts were 130μ by 95μ in size and contained about a hundred sporocysts. Between the sporocysts were a number of round refractile bodies up to 8μ in diameter. The sporocysts varied in size from 21·4μ to 36·4μ in length by 15·7μ to 20μ in breadth and contained four to sixteen sporozoites and a residual body, from 3·6μ to 6·4μ in diameter, enclosed in a definite membrane. Apart from the residual body each sporocyst contained a number of small refractile granules lying apparently in the sporozoites.

In the sporocysts the sporozoites were seen to be actively motile, in some cases sufficiently so to cause the whole sporocyst to spin.

From the ruptured sporocysts sporozoites were seen emerging; each sporozoite was then observed to be lying in a membrane which when released from the sporocyst assumed the form of an elongated

spindle about 35μ in length and 6·4μ in breadth (figs. 6 to 8, and 12). The small refractile granules noted in the sporocyst were found to lie in the membrane outside the sporozoite, each membrane containing two to four granules.

The membranes, including the sporozoites, were all seen to be divided longitudinally by two fine lines (fig. 12). The sporozoites were actively motile within their membranes, constantly changing their shape and size by a series of contractile movements, so that it is difficult to give definite measurements. When fully stretched out the sporozoites were sickle-shaped with one end pointed and the other blunt, and then measured 34μ in length by 5μ in their thickest part (fig. 9). Each sporozoite contained a round nucleus.

By contracting and thus increasing their transverse diameter the sporozoites stretched the enclosing membrane and created a gap between the two longitudinal lines of the membrane (fig. 8). The sporozoites then slowly worked their way through the gap, leaving an empty husk containing several refractile granules (fig. 12).

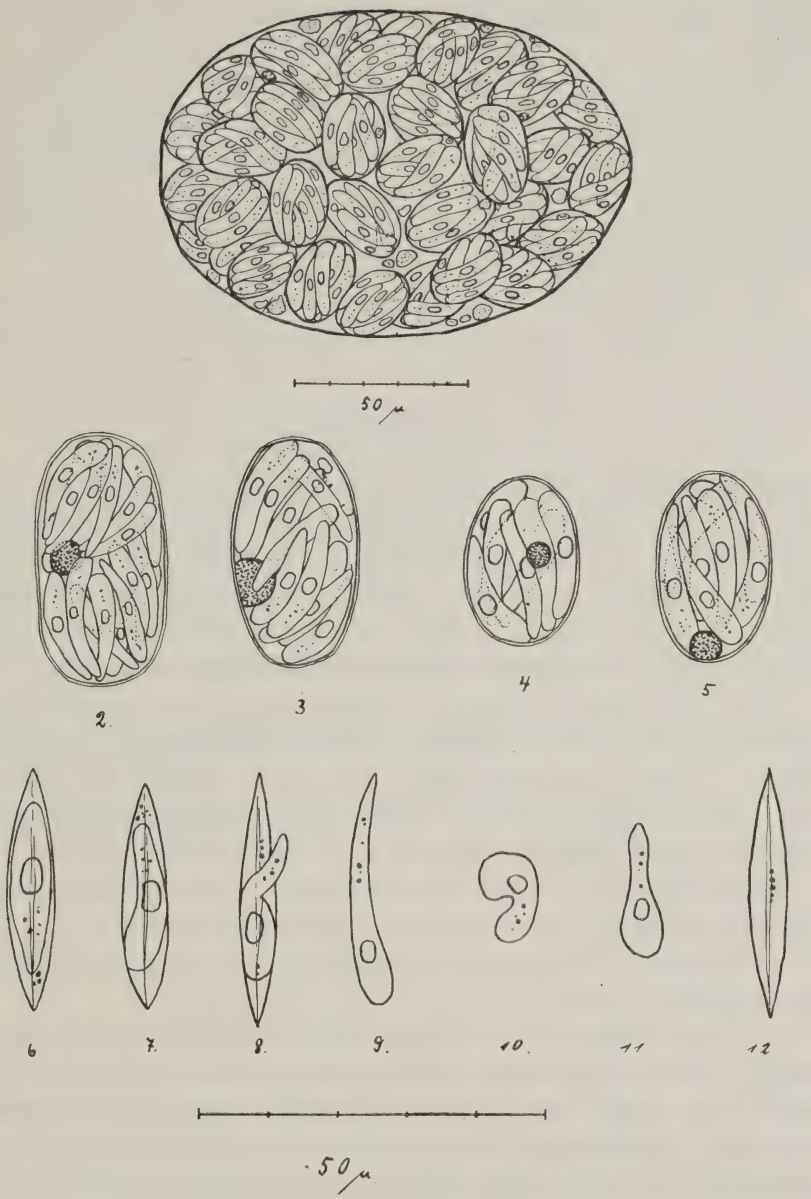
Having escaped, the sporozoites continue their contractile movements, constantly changing their shape and, at the same time, performing a slow translatory movement.

All the material was transferred to two slides and examined in the fresh.

As it seemed obvious that the oocyst above described formed a part of the life-cycle of a haemogregarine of a vertebrate, the following experiments were immediately performed.

(i) The contents of one fresh preparation containing intact sporocysts and numerous free sporozoites from ruptured sporocysts were carefully washed off the slide in 0·5 c.c. normal saline ; 0·25 c.c. of the resulting mixture was injected intraperitoneally into a specimen of *Gongylus ocellatus*, and the remainder intraperitoneally into a specimen of *Mabuia quinquefasciata*. The above two lizards were both free from haemogregarines at the time of the experiment. The gecko *Hemidactylus turcicus*, which is common in houses in Jericho and feeds on sandflies, would have been a more suitable animal for the experiment, but unfortunately no specimen of this animal was at the moment available in the laboratory.

(ii) The material from the second fresh preparation containing numerous sporocysts and free sporozoites was rubbed into puncture



FIGS. 1-12.

1. A complete oocyst.
2-5. Sporocysts.
6-7. Sporozoites in their enclosing membrane.
8. Sporozoite escaping from membrane.
9-11. Change in shape of a sporozoite.
12. Empty membrane with refractile granules.

wounds made by a needle into the forearms of two healthy human beings.

The lizards were examined at intervals during an observation period of five weeks and the peripheral blood was found to contain no haemogregarines. At the end of this period the two animals were killed and the liver, lungs and bone marrow were examined for schizonts with a negative result. The blood of the two human beings was also found to be negative during this period.

The oocyst described above apparently belongs to the genus *Hepatozoon* (Miller 1908) since it contains numerous sporocysts and each sporocyst contains a number of sporozoites, the formula for the genus *Hepatozoon* being, according to Reichenow (1921) :

' Oocysts with *n* sporocysts, sporocysts with *n* sporozoites.'

Infection of a new vertebrate host with *Hepatozoon* sp. takes place by the accidental ingestion of the transmitting arthropod containing ripe oocysts, e.g., *Mus rattus* becomes infected with *Hepatozoon perniciosum* (Miller 1908) by swallowing the mite *Lelaps echidnius* containing ripe oocysts, and dogs are infected with *Hepatozoon canis* (James 1905) by swallowing infected *Rhipicephalus sanguineus*.

Up to the present the genus *Hepatozoon* has been recorded only from mammals, but the finding of oocysts belonging to the genus *Hepatozoon* free in the abdominal cavity of *Gl. palpalis* by Chatton and Roubaud (1913) and by Macfie (1916) points to the possible presence of this genus in lizards or birds which feed on the fly.

In the case of the oocyst of *Phlebotomus papatasii*, two possible modes of infecting a vertebrate host suggest themselves :

1. Ingestion by a lizard and liberation of the sporozoites into the alimentary canal.

2. The crushing of an infected Phlebotomus during the act of feeding, and the liberation of sporozoites into the wound, i.e., the method by which Phlebotomus is generally assumed to transmit cutaneous leishmaniasis. The experiment on two human beings described above thus approximates to an infection under natural conditions.

A number of observers (Krempf 1917, Dimond 1917, Sergent, Et. and Ed. and Parrot 1922, Noc 1922, Nattan-Larrier 1922), have described haemogregarines from man. The findings of all these authors have been subjected to a destructive criticism by

Wenyon (1923), who concluded that ' the haemogregarines of man have still to be found.'

The bionomics of *P. papatasii* render it an eminently suitable transmitting agent of a haemogregarine to man if such occur and the result of the experiment on two human beings is therefore of interest.

The above is the first record of an oocyst in *Phlebotomus papatasii*.

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